

Twin-sibling study and meta-analysis on the heritability of maximal oxygen consumption

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Schutte NM, Nederend I, Hudziak JJ, Bartels M, de Geus EJC. Twin-sibling study and meta-analysis on the heritability of maximal oxygen consumption. *Physiol Genomics* 48: 210–219, 2016. First published January 19, 2016; doi:10.1152/physiolgenomics.00117.2015.—Large individual differences exist in aerobic fitness in childhood and adolescence, but the relative contribution of genetic factors to this variation remains to be established. In a sample of adolescent twins and siblings ($n = 479$), heart rate (HR) and maximal oxygen uptake ($\dot{V}O_{2\max}$) were recorded during the climax of a graded maximal exercise test. In addition, $\dot{V}O_{2\max}$ was predicted in two graded submaximal exercise tests on the cycle ergometer and the treadmill, using extrapolation of the HR/ $\dot{V}O_2$ curve to the predicted HR_{max}. Heritability estimates for measured $\dot{V}O_{2\max}$ were 60% in ml/min and 55% for $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹. Phenotypic correlations between measured $\dot{V}O_{2\max}$ and predicted $\dot{V}O_{2\max}$ from either submaximal treadmill or cycle ergometer tests were modest ($0.57 < r < 0.70$), in part because of the poor agreement between predicted and actual HR_{max}. The majority of this correlation was explained by genetic factors; therefore, the submaximal exercise tests still led to very comparable estimates of heritability of $\dot{V}O_{2\max}$. To arrive at a robust estimate for the heritability of $\dot{V}O_{2\max}$ in children to young adults, a sample size weighted meta-analysis was performed on all extant twin and sibling studies in this age range. Eight studies, including the current study, were meta-analyzed and resulted in a weighted heritability estimate of 59% (ml/min) and 72% (ml·min⁻¹·kg⁻¹) for $\dot{V}O_{2\max}$. Taken together, the twin-sibling study and meta-analyses showed that from childhood to early adulthood genetic factors determine more than half of the individual differences in $\dot{V}O_{2\max}$.

maximal exercise test; $\dot{V}O_{2\max}$; adolescents; genetics

MAXIMAL OXYGEN UPTAKE ($\dot{V}O_{2\max}$) is defined as the highest rate of oxygen consumption during maximal intensity exercise performed until exhaustion (23) and is considered a good index of aerobic fitness and endurance capacity. Direct measurement of oxygen consumption and carbon dioxide production during the climax of a graded maximal exercise test is the gold standard $\dot{V}O_{2\max}$ measurement. Large individual differences exist in maximal exercise test-derived $\dot{V}O_{2\max}$, and, although these are significantly correlated to the regular exercise status of a subject, this correlation is not as strong as generally assumed. Various measures of total physical activity or regular leisure time sports and exercise behavior generally show only modest association with $\dot{V}O_{2\max}$ (1, 6, 38, 43). The variation in baseline $\dot{V}O_{2\max}$ in sedentary subjects is often already much

larger than the training-induced increase over this baseline, which is on average only about 25% (13, 20, 34, 46). Training furthermore increases rather than decreases the individual differences seen at baseline, as the $\dot{V}O_{2\max}$ response to training itself shows large variation (10, 39).

The above pattern suggests an important role for innate factors in the population variation in $\dot{V}O_{2\max}$, and twin and family studies seem to confirm this (8, 9, 17, 24, 25, 27, 28, 30, 32, 33, 42). Table 1 provides an overview of correlations among relatives i.e., monozygotic (identical, MZ) and dizygotic (fraternal, DZ) twins, siblings, and parents with their offspring. The monozygotic twin correlations in Table 1 range from 0.62 to 0.95. The DZ twin correlations, sibling correlations, and parent-offspring correlations also vary substantially across studies but are systematically lower than the MZ twin correlations. In line with the variability in twin correlations, heritability estimates have varied widely. Possible sources of this variation are differences in the age of the participants, differential approaches to adjustment for body mass and/or body composition, training status of the subjects, or differences in protocol or fitness equipment (i.e., cycle ergometer or treadmill) that was used to measure or predict $\dot{V}O_{2\max}$ between the various studies. A major source, however, seems to be the rather modest sample sizes. As is clear from Table 1 there are only two studies with large samples (28, 42), but both these larger studies used a submaximal instead of a maximal exercise test. These tests do not measure $\dot{V}O_{2\max}$ directly but predict it from an exercise test that is halted at a predetermined point [certain percent of the predicted maximal heart rate (HR_{max})] below the maximal exercise capability of the individual. Since it does not demand $\dot{V}O_{2\max}$ measurement during exhaustive exercise, the submaximal exercise test is better suited in larger (genetic) epidemiological studies. However, it is currently unknown whether a submaximal exercise test correctly captures the genetic factors influencing $\dot{V}O_{2\max}$.

One of the most used submaximal exercise test is the nomogram of Åstrand, which requires cycling on a constant individually chosen work rate. $\dot{V}O_{2\max}$ is predicted using the steady-state heart rate (HR) achieved after 6 min (3). This method has clear limitations as results may be influenced by individual differences in submaximal HR at a given work rate due to training status, resting HR, and body composition. Estimated $\dot{V}O_{2\max}$ with this method showed correlations in the range of 0.47 and 0.82 with measured $\dot{V}O_{2\max}$ in adult populations (14, 16, 21, 22, 37). More promising is the $\dot{V}O_{2\max}$ prediction using a graded submaximal exercise protocol in which the intensity increases at regular intervals up to but never exceeding a certain percent of the HR_{max}. $\dot{V}O_{2\max}$ can be

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Table 1. Overview of genetic studies on $\dot{V}O_{2\max}$ conducted in a twin and/or family design

Study	Subjects	$\dot{V}O_{2\max}$ Measurements	r_{MZ}	r_{DZ}	$r_{sibling}$	$r_{parent-offspring}$	Heritability
Klissouras et al., 1971	15 MZ 10 DZ age: 10 ± 2 yr	maximal exercise test treadmill ml/min	0.91	0.44			93%
Klissouras et al., 1973	23 MZ 16 DZ age: 27 ± 14 yr	maximal exercise test treadmill ml·min ⁻¹ ·kg ⁻¹	0.95	0.36			
Montoye & Gayle 1978	93 father-son pairs 70 brother pairs age: 10–69 yr	<39 yr maximal exercise test >39 yr submaximal exercise test (until HR = 160) treadmill l/min corrected for age, weight, skinfolds			0.18	0.34	
Lortie et al., 1982	594 parent-offspring pairs 223 sibling pairs age: 42 ± 4 yr (parents) age: 15 ± 3 yr (children)	submaximal exercise test (until HR = 150/170) treadmill ml/min ml·min ⁻¹ ·kg ⁻¹ $\dot{V}O_{2\max}$ predicted corrected for sex, age, skinfolds, physical activity, cigarette smoking, and SES			0.36 ^a 0.33 ^b	0.21 ^a 0.17 ^b	
Lesage et al., 1985	96 parent-offspring pairs 39 sibling pairs age: 43 ± 5 yr (parents) age: 16 ± 4 yr (children)	maximal exercise test treadmill ml/min ml·min ⁻¹ ·kg ⁻¹ corrected for sex and age			0.14 ^a 0.19 ^b	0.06 ^a 0.03 ^b	
Bouchard et al., 1986	106 MZ 66 DZ 27 sibling pairs age: 22 ± 3 yr	maximal exercise test cycle ergometer ml·min ⁻¹ ·kg ⁻¹ corrected for sex and age	0.70	0.51	0.41		38%
Fagard et al., 1991	29 MZ 19 DZ age: 22 ± 4 yr	maximal exercise test cycle ergometer ml/min ml·min ⁻¹ ·kg ⁻¹	0.77 ^a 0.77 ^b	0.05 ^a 0.04 ^b			77% 68%
Sundet et al., 1994	436 MZ 622 DZ age: late teens/early 20s	only males, restricted age range submaximal exercise test (until HR = 140) cycle ergometer ml·min ⁻¹ ·kg ⁻¹ $\dot{V}O_{2\max}$ predicted ^c	0.62	0.29			62%
Maes et al., 1996	43 MZ 61 DZ 84 fathers 97 mothers age: 39 ± 4 yr (parents) age: 10 yr (children)	maximal exercise test treadmill l/min restricted age range	0.75	0.32		0.25/0.31 ^d	69%/87% ^e
Bouchard et al., 1998	125 sons 134 daughters 85 fathers 85 mothers age: 52 ± 5 yr (parents) age: 25 ± 6 (children)	maximal exercise test cycle ergometer ml/min corrected for sex and age			0.36	0.14/0.36	59%
Mustelin et al., 2011	59 MZ 92 DZ age: 27 ± 2 yr	maximal exercise test cycle ergometer ml/min corrected for sex, restricted age range	0.64	0.21			65%

^a r_{MZ} , Monozygotic twin correlation; ^b r_{DZ} , dizygotic twin correlation; ^c $r_{sibling}$, sibling correlation; ^d $r_{parent-offspring}$, parent-offspring correlation; ^aml/min; ^bml·min⁻¹·kg⁻¹; ^cpredicted $\dot{V}O_{2\max}$ was transformed to a categorical score from 1 to 9. The correlations are based upon those categorical scores; ^dfather-child correlation/mother-child correlation; ^eheritability estimate for males/heritability estimate for females. HR, heart rate; SES, socioeconomic status.

obtained by extrapolating the HR/ $\dot{V}O_2$ curve to the predicted HR_{max}, allowing for individual differences in $\dot{V}O_2$ /HR slope. This estimation method showed correlations in the range of 0.76 and 0.98 with measured $\dot{V}O_{2\max}$ in adult populations (16, 19, 26), although it is sensitive to the protocol used. Submaximal tests on a cycle ergometer yield lower predicted $\dot{V}O_{2\max}$ values than tests on a treadmill (19, 31).

Adolescent $\dot{V}O_{2\max}$ has been measured in parent-offspring studies using submaximal exercise tests (27, 28), but a striking omission in Table 1 is adolescent twin studies using a maximal

exercise test to examine $\dot{V}O_{2\max}$ in an adolescent population. The aim of the current study is to address this gap in the extant literature. In a large sample of adolescent twins and siblings, HR and $\dot{V}O_2$ were recorded during the climax of a graded maximal exercise test. $\dot{V}O_{2\max}$ was further predicted from two graded submaximal exercise tests on the cycle ergometer and the treadmill, using extrapolation of the HR/ $\dot{V}O_2$ curve to the predicted HR_{max}. This allowed us to address our second aim: to test the extent to which the genetic factors influencing measured $\dot{V}O_{2\max}$ during a maximal exercise test overlap with those

influencing predicted $\dot{V}O_{2\max}$ from submaximal exercise tests. Information on the genetic overlap between measured and predicted $\dot{V}O_{2\max}$ can reveal whether they can be used interchangeably in genetic association studies aiming to identify the genetic variants underlying $\dot{V}O_{2\max}$. A high degree of overlap would mean that submaximal exercise tests, which are easier to implement in large-scale genetic studies, might suffice for such studies. Twin correlations, heritability of the measured and predicted $\dot{V}O_{2\max}$, as well as the genetic covariance among these parameters were estimated in a multivariate design. We hypothesize that a substantial part of the variation in $\dot{V}O_{2\max}$ in our adolescent sample is explained by genetic factors. As previous studies in adults showed high correlations between $\dot{V}O_{2\max}$ predicted from a graded submaximal exercise protocol and measured $\dot{V}O_{2\max}$, we expect moderate to high phenotypic correlations and a significant contribution of genes to this correlation. Finally, a sample size weighted meta-analysis was performed on the univariate twin correlations obtained from all twin studies in the age range of 10–30 yr (including the current study) that measured $\dot{V}O_{2\max}$, aiming to arrive at a more robust estimate for the heritability of this crucial trait in exercise physiology.

MATERIALS AND METHODS

Sample

Healthy adolescent twin pairs aged between 16 and 18 yr and their siblings (age range 12–25 yr) from the Netherlands Twin Register (44) were invited to participate in a study on the determinants of adolescent exercise behavior. Selection for invitation was based on the availability of longitudinal survey data on zygosity and regular leisure time exercise behavior. The aim was to have sufficient individuals present from the entire spectrum of sedentary to vigorous leisure time exerciser and for each zygosity group. We started with a random selection, but if a zygosity group was underrepresented or if there were too few sedentary or vigorous exercisers, invitations were biased toward the underrepresented groups. This was mainly the case for sedentary subjects; twins who reported no engagement in exercise behavior on a previously filled out survey were selected for invitation. The cotwin was then selected as well, regardless of her or his exercise status. To be eligible for the study, participants had to have no history of cardiovascular or respiratory disease and be physically capable of engaging in exercise activities.

Participants were invited by letter advertising the opportunity to test their fitness in addition to earning a gift voucher. All invitees had to be able and willing to visit the Vrije Universiteit in Amsterdam for lab testing. For the current study, a complete dataset was available for 479 subjects: 221 complete twin pairs: 112 MZ pairs and 109 DZ pairs and 33 of their singleton siblings. In addition, two nontwin sibling pairs participated. This sample size should be sufficient to detect univariate genetic influences with a power of 80% (assuming substantial heritability estimates of 60%, based on previous studies) (35).

All participants provided written informed consent, and if the participants were under 18 yr, consent was given by both of their parents/guardians. All study procedures submitted to and approved by the Medical Ethics Review Committee of the VU University Medical Center Amsterdam (NL35634.029.10).

Procedure

On participants' arrival at the laboratory, height and weight were measured and a short lifestyle interview was completed, including detailed questions on current levels of regular exercise. Next, two exercise test were conducted (in fixed order) on a electromechanically

braked Lode cycle ergometer (type Corival) and a Lode treadmill (type Valiant) at fixed loads that are below the intensity of the ventilatory threshold for most adolescents.

The first session on the cycle ergometer started with a 2 min warming-up period, followed by four incremental stages of 5 min each [males: 70, 90, 110, 130 Watts (W); females: 40, 60, 80, 100 W]. Participants were instructed to pedal at a fixed rounds per minute (rpm): between 60 and 70 rpm. The test ended with a 1 min cooling-down phase, followed by a 5 min recovery period. The second session on the treadmill consisted of a 1 min warm-up period, followed by four incremental stages of 5 min each (males: 6, 6.5, 7 and 8 km/h; females: 5.5, 6, 6.5 and 7 km/h). Again, the test ended with a 1 min cooling-down phase, followed by a 5 min recovery period. To ensure that the intensity of every stage was below the intensity of the ventilatory threshold for most adolescents, the ratio of the oxygen consumption and carbon dioxide production ($\dot{V}O_2/\dot{V}CO_2$) was monitored. This respiratory exchange ratio (RER) can be used to estimate the ventilatory threshold (41). This threshold is transcended when exhalation of CO_2 exceeds inhalation of O_2 , which is visualized by a $RER > 1.00$. For each test the load of each stage was adjusted when necessary to keep the intensity below an RER of 0.95.

Finally, an incremental maximal exercise test was conducted on the cycle ergometer to establish $\dot{V}O_{2\max}$. The work rate was increased every minute until exhaustion while participants pedaled at 60–100 rpm. In the standard protocol male started at 75 W with increments of 25 W/min. For females stage one started at 70 W and work load was increased by 20 W/min. Adjustments to this protocol (higher increasing work load every step) were done by experienced researchers based on the exercise behavior, age, height, and weight of the participant. The test was terminated when the participant was not able to keep rpm above 50 despite serious attempts. After cessation of the test, every participant completed a mandatory cool-down phase on the cycle ergometer of 5 min on a low, individually chosen work rate.

Measurements

Regular exercise behavior. Leisure time exercise behavior was measured by a short lifestyle interview, in which the participants indicated what types of regular sports or exercise activities they were involved in. Subjects were asked to indicate for each activity for how many years the subject participated in the activity, for how many months a year, how many times a week, and how many minutes each time. Each activity was recoded into a metabolic equivalent (MET) score, based on the compendium of energy expenditure (2). By multiplying the MET score, the frequency, and the duration of each exercise activity, we calculated weekly MET-hours spent on exercise activities for each participant. We only included activities that were conducted for at least 3 mo a year and more than half a year previously (thereby excluding ski holidays, sailing camps, and similar). In addition, subjects were asked to indicate how much time per week was spent on physical activity related to active transportation (walking, cycling) and compulsory physical education classes, but MET-hours spent on these activities were kept separate and not used in our index of voluntary exercise behavior in leisure time.

Gas exchange. $\dot{V}O_2$ and carbon dioxide production ($\dot{V}CO_2$) were recorded breath-by-breath by means of a telemetric gas exchange system (Cosmed K4b², Rome, Italy). During the course of the experiment, the main sample unit and the battery pack were attached to the back of the subject. Before each test, the O_2/CO_2 analysis system was calibrated with ambient air and a gas mixture that had an O_2 concentration of 16% and a CO_2 concentration of 5%. The calibration of the turbine flowmeter was performed by via a 3 l syringe (all according to the manufacturer's instructions). Figure 1 illustrates the changes in $\dot{V}CO_2$ and $\dot{V}O_2$ across the entire experimental protocol for a pair of MZ and a pair of DZ twins.

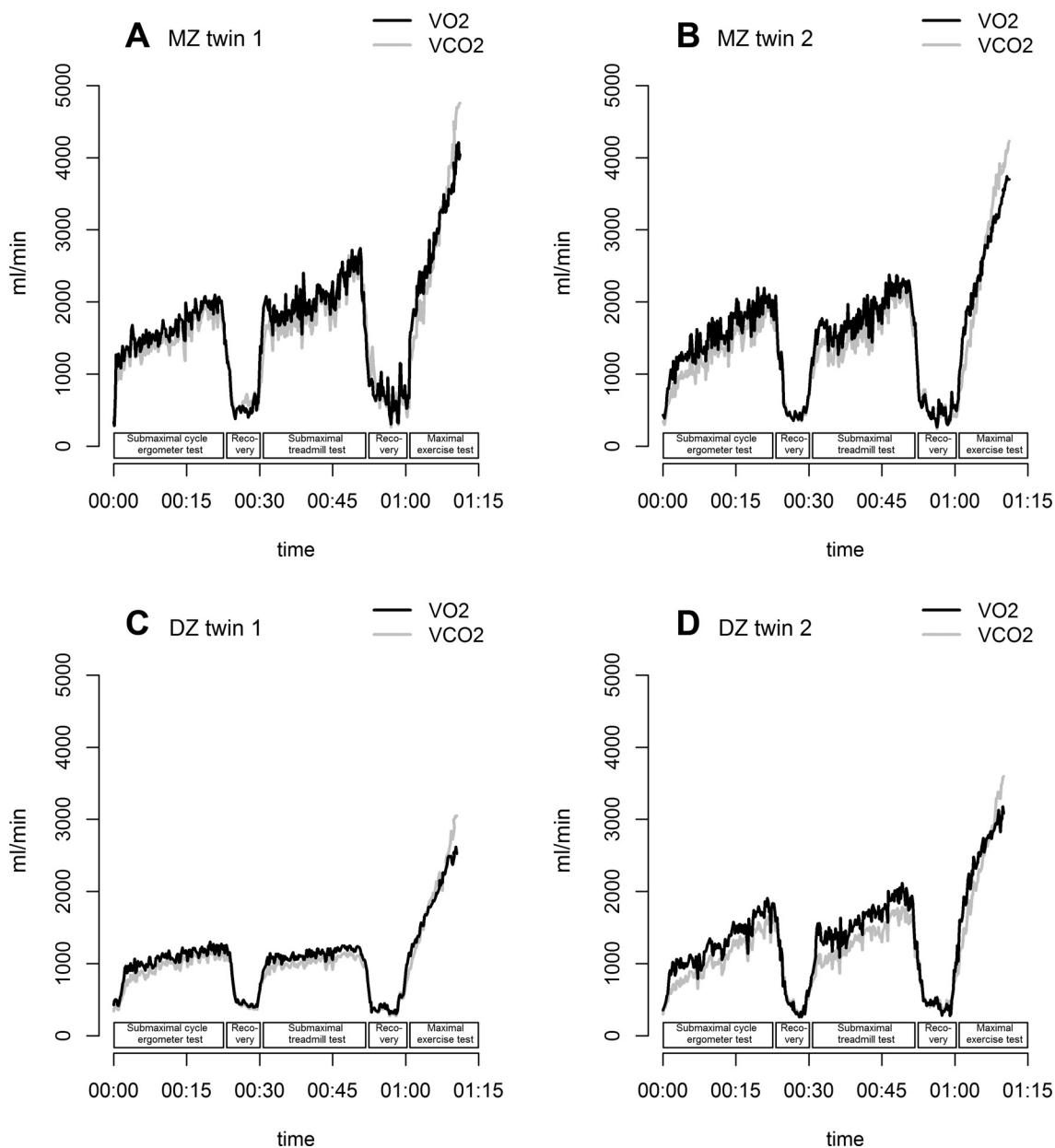


Fig. 1. Changes in oxygen uptake ($\dot{V}O_2$) and carbon dioxide production ($\dot{V}CO_2$) across the entire experimental protocol for a pair of monozygotic (MZ) (A and B) and a pair of dizygotic (DZ) twins (C and D). The 2 submaximal exercise tests on the cycle ergometer and treadmill and the final maximal exercise test are clearly visible as $\dot{V}O_2$ and $\dot{V}CO_2$ increase when subjects start exercising. The MZ twins resemble each other more than DZ twins in absolute $\dot{V}O_2$ and $\dot{V}CO_2$.

HR. The electrocardiogram (ECG) was recorded continuously with the VU-AMS5fs device (VU University, Amsterdam, the Netherlands). This device was developed to study autonomic nervous system activity in naturalistic settings (15). The version used here measured the ECG together with the impedance cardiogram from five disposable, pregelled Ag/AgCl electrodes. Due to the portable nature of this device, the participants were not discomforted by wearing this on the hip during the exercise tests. HR was obtained from the ECG by an automated R-wave peak detector in the VU-AMS software suite (VU-DAMS version 3.1; Vrije Universiteit, Amsterdam, the Netherlands, <http://www.vu-ams.nl>) and shown online during testing. Data analysis was based on automated offline scoring of the R-waves, with suspicious interbeat intervals (too short or too long taken the local mean and variance) corrected by interpolation or excluded by marking these beats as artifacts during visual inspection of the ECG signal.

Data Processing

Measuring $\dot{V}O_{2\max}$ during maximal exercise. To obtain $\dot{V}O_{2\max}$, only $\dot{V}O_2$ data with a corresponding RER of at least 1.10 were selected to ensure good effort above the intensity of the ventilatory threshold. Breath-by-breath $\dot{V}O_2$ data was cut into 20 s blocks. For every 20 s block, we calculated the mean $\dot{V}O_2$, after discarding deviant breaths. $\dot{V}O_{2\max}$ was determined as the highest mean value of $\dot{V}O_2$ of all the 20 s blocks. The HR_{\max} in that specific block was taken as corresponding HR_{\max} .

Predicting $\dot{V}O_{2\max}$ from submaximal exercise. To predict $\dot{V}O_{2\max}$, breath-by-breath $\dot{V}O_2$ data and beat-to-beat HR data were synchronized and the mean of every 5 s block was calculated for submaximal cycle and treadmill exercise tests separately. Using the univariate regression function in SPSS (IBM SPSS Statistics for Windows, version 20.0; IBM, Armonk, NY), we examined the relationship

between $\dot{V}O_2$ (dependent variable) and HR (independent variable) and calculated a slope and intercept for every subject for the submaximal cycle ergometer test as well as for the submaximal treadmill test. Using these parameter estimates together with HR_{\max} we calculated the predicted $\dot{V}O_{2\max}$ for every subject. Because we wanted to test the feasibility of using submaximal tests only, HR_{\max} was obtained from the formula $208 - 0.7 \cdot \text{age}$ (43) rather than the actual measured HR_{\max} , although analyses were repeated using the actual measured HR_{\max} .

Genetic Analyses

Genetic structural equation modeling was done in OpenMx (5) under R (R Development Core Team, 2011) with the raw-data ML procedure for estimation of parameters. For all analyses, a threshold of $P < 0.05$ was considered for statistical significance. All $\dot{V}O_{2\max}$ values were Z-transformed. Since (nontwin) siblings share, like DZ twins, on average 50% of their genes, parameter estimates were constrained to be equal for DZ twins and siblings. First, a trivariate model that estimated all parameters freely (a saturated model) was fitted, including the measured $\dot{V}O_{2\max}$ and the $\dot{V}O_{2\max}$ predicted from the submaximal cycle and treadmill test. Main effects of sex and age on mean levels of these phenotypes were considered in the model. We tested the significance of these covariates by comparing the model, including the specific component, with a model in which the component is constrained to be equal to zero. These nested submodels were compared by hierarchic χ^2 tests. The χ^2 statistic is computed by subtracting log-likelihood ($-2LL$) for a reduced model from the $-2LL$ for the full model ($\chi^2 = -2LL_{\text{full model}} - -2LL_{\text{reduced model}}$). This χ^2 statistic is distributed with degrees of freedom (df) equal to the difference in the number of parameters estimated in the two models ($\Delta df = df_{\text{full model}} - df_{\text{reduced model}}$). If the difference test is significant, the constraints on the reduced model cause a significant deterioration of the fit of model. Twin and cross-twin/cross-trait correlations and their 95% confidence intervals (CI) were estimated for the MZ and DZ twins/siblings.

Subsequently, a trivariate Cholesky decomposition was fitted to the data. This decomposition model decomposes the total phenotypic variance into sources of additive genetic variance/covariance (A), dominant genetic variance/covariance (D) or shared (familial) environmental variance/covariance (C) and unique environmental variance/covariance (E). The C and D effects cannot be estimated simultaneously in a twin/sibling model. Therefore, the ratio of the MZ correlations to the DZ/sibling correlations was used to determine which model (ACE or ADE) is most appropriate. We tested the significance of the variance-covariance components by comparing the model including the specific component to a model in which the component is constrained to be equal to zero.

Meta-analysis

A search of the electronic databases ISI Web of Knowledge and PubMed was conducted with the key words: maximal oxygen uptake, $\dot{V}O_{2\max}$, aerobic capacity, aerobic performance, cardiorespiratory (fitness) and genes, heritability, twin(s), family (date last searched: January 2015). Furthermore, the reference lists of these articles were inspected. Articles published in English reporting twin, sibling, and/or parent-offspring correlations and corresponding sample sizes and with participants with an age < 30 yr were selected. Only articles in which $\dot{V}O_2$ was measured in a maximal exercise protocol or predicted from a submaximal exercise protocol were included. All twin and sibling correlations of these articles (including the current study) were included in a sample size weighted meta-analysis for $\dot{V}O_{2\max}$ expressed in ml/min and $\dot{V}O_{2\max}$ expressed in $\text{ml} \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$. Twin correlations from the current study were calculated in univariate models without the siblings to be comparable with the twin correlations included in the meta-analysis.

Table 2. Means and SD of measured and predicted $\dot{V}O_{2\max}$ in ml/min and $\text{ml} \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$, measured and predicted HR_{\max} and minutes per week spent on walking and cycling (transportation), physical education class, and leisure time exercise behavior in METs of males and females

	Males (n = 233)		Females (n = 230)	
	Mean	SD	Mean	SD
Body composition				
Height, cm	180.4	7.8	168.3	6.6
Weight, cm	67.3	10.3	61.8	9.7
BMI, kg/m	20.6	2.5	21.8	3.3
$\dot{V}O_{2\max}$, ml/min	3,132	540	2,240	316
Measured				
Predicted from cycle ergometer test				
	2,933	648	2,021	389
Predicted from treadmill test				
	2,968	606	2,029	400
$\dot{V}O_{2\max}$, $\text{ml} \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$				
Measured				
	46.9	6.9	36.7	5.6
Predicted from cycle ergometer test				
	43.8	8.1	33.0	6.0
Predicted from treadmill test				
	44.5	8.1	34.1	6.3
Heart rate, bpm				
Resting heart rate				
	72.6	11.4	75.6	11.2
Maximal heart rate measured				
	195.4	10.1	195.2	8.9
Maximal heart rate predicted (Tanaka)				
	196.1	0.8	195.9	0.9
Regular exercise				
Walking, min/wk				
	38.9	71.8	43.5	79.8
Cycling, min/wk				
	233.1	156.3	209.4	163.3
Physical education, min/wk				
	151.6	139.9	132.4	112.4
Leisure-time exercise, METs/wk				
	25.7	22.5	19.2	22.1

MET, metabolic equivalent; BMI, body mass index.

In OpenMx, a variance decomposition model was fitted to the twin correlations (weighted for sample size) to estimate the influence of additive genetic (A) and shared environmental influences (C) on $\dot{V}O_{2\max}$ in ml/min and $\dot{V}O_{2\max}$ in $\text{ml} \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$ according to the approach of Bartels et al. (4). First, the twin and sibling correlations were used to estimate the genetic and environmental influences for each study separately. Subsequently, all studies were taken together to estimate one weighted heritability estimate for $\dot{V}O_{2\max}$. These two models were compared by the hierarchic χ^2 test. A significant deterioration of the fit of model indicated significant heterogeneity across the studies (4). We repeated the meta-analysis by excluding the study by Sundet et al. (42), which used predicted $\dot{V}O_{2\max}$ from submaximal exercise testing to also provide a weighted heritability estimate of actual measured $\dot{V}O_{2\max}$.

RESULTS

General Descriptives

Means and standard deviations for measured $\dot{V}O_{2\max}$ and $\dot{V}O_{2\max}$ predicted from the submaximal cycle and treadmill test and measured and predicted HR_{\max} of males and females are shown in Table 2. Fifteen subjects did not meet the RER > 1.10 criterion. For nine of these subjects there was no sufficient evidence that they exercised until exhaustion according to the experimental researcher report and/or the HR_{\max} was $< 85\%$ of HR_{\max} . Therefore, these nine subjects and their coinciding twin/sibling were excluded from further analyses involving measured $\dot{V}O_{2\max}$. The final sample size consisted of 463 subjects. Means and standard deviations of minutes spent on walking, cycling, physical education class, and MET scores for

leisure time exercise behavior are presented in Table 2. Although the age range is small, significant age effects ($\dot{V}O_{2\max}$ increases with age) were found on $\dot{V}O_{2\max}$ in ml/min ($P < 0.001$), but not for $\dot{V}O_{2\max}$ expressed in $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$. Both predicted and measured $\dot{V}O_{2\max}$ are higher in males than in females (all $P < 0.001$). Furthermore, males were more engaged in weekly exercise behavior ($P = 0.002$). As expected, weekly leisure time exercise behavior correlated significantly with $\dot{V}O_{2\max}$, but the correlation was modest: $r = 0.28$ with $\dot{V}O_{2\max}$ in ml/min and $r = 0.34$ (both $P < 0.001$) with $\dot{V}O_{2\max}$ $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$. The correlation between measured $\dot{V}O_{2\max}$ and weekly minutes of cycling is significant ($r = 0.25$ for $\dot{V}O_{2\max}$ expressed in ml/min and $r = 0.22$ for $\dot{V}O_{2\max}$ expressed in $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$, both $P < 0.001$), whereas the correlations between measured $\dot{V}O_{2\max}$ and weekly minutes of walking or weekly hours of physical education class are small ($-0.12 < r < 0.03$).

Correlation between Measured and Predicted $\dot{V}O_{2\max}$

Measured $\dot{V}O_{2\max}$ in ml/min showed a correlation of 0.70 (95% CI: 0.65–0.75) with $\dot{V}O_{2\max}$ predicted from the submaximal cycle test and 0.64 (95% CI: 0.58–0.70) with $\dot{V}O_{2\max}$

predicted from the treadmill test. Likewise, measured $\dot{V}O_{2\max}$ in $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$ was significantly correlated with $\dot{V}O_{2\max}$ predicted from the submaximal cycle test ($r = 0.61$, 95% CI: 0.55–0.68) and with $\dot{V}O_{2\max}$ predicted from the submaximal treadmill test ($r = 0.57$, 95% CI: 0.50–0.64). Despite the significant relationship between predicted and measured $\dot{V}O_{2\max}$, Bland Altman plots in Fig. 2 show considerably discrepancy between these measures, expressed in ml/min. Regression of the mean of the two measurements (measured and predicted $\dot{V}O_{2\max}$) on the difference between the two values (y-axis) shows that the discrepancy increases as absolute $\dot{V}O_{2\max}$ increases. In males the absolute differences in measured and predicted $\dot{V}O_{2\max}$ are larger than in females. A potential source of error is the use of an age-predicted HR_{\max} . Absolute mean differences between measured (195 ± 10) and predicted HR_{\max} (202 ± 1) are greater than zero ($p < 0.001$). Repeating the analyses with measured HR_{\max} significantly improved the correlation of measured to predicted $\dot{V}O_{2\max}$ from the submaximal cycle test (in ml/min $r = 0.76$, 95% CI: 0.72–0.90; in $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$ $r = 0.69$, 95% CI: 0.64–0.74) and to predicted $\dot{V}O_{2\max}$ from the treadmill test (in ml/min $r = 0.71$, 95% CI: 0.66–0.76; in $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$ $r = 0.65$, 95% CI: 0.59–0.70).

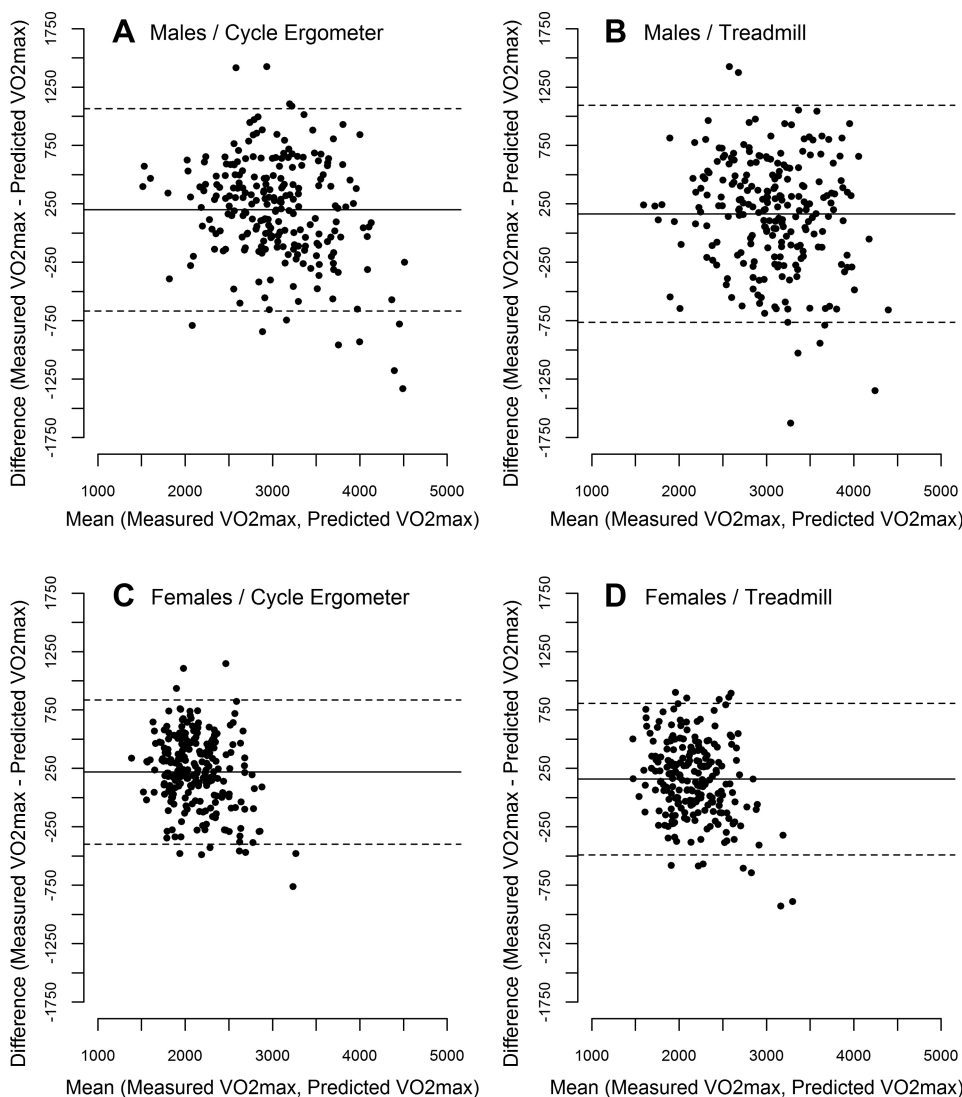


Fig. 2. Bland-Altman plots for maximal oxygen uptake ($\dot{V}O_{2\max}$) in ml/min. The x-axis shows the mean of the 2 measurements (measured and predicted $\dot{V}O_{2\max}$), and the y-axis shows the difference between the 2 values. The solid line represents the mean difference. The dotted lines represent the average difference ± 1.96 SD of the difference. A: male $\dot{V}O_{2\max}$ submaximal cycle test; B: male $\dot{V}O_{2\max}$ submaximal treadmill test; C: female $\dot{V}O_{2\max}$ submaximal cycle test; D: female $\dot{V}O_{2\max}$ submaximal treadmill test.

Table 3. Twin (diagonal) and cross-twin/cross-trait (off diagonal) correlations (95% CI) estimated from the saturated model for measured $\dot{V}O_{2\max}$ and $\dot{V}O_{2\max}$ predicted from the submaximal cycle and treadmill tests

	$\dot{V}O_{2\max}$, ml/min			$\dot{V}O_{2\max}$, ml·min ⁻¹ ·kg ⁻¹		
	Measured	Predicted from Cycle Test	Predicted from Treadmill Test	Measured	Predicted from Cycle Test	Predicted from Treadmill Test
<i>MZ correlations</i>						
Measured	0.61 (0.50, 0.70)			0.53 (0.40, 0.63)		
Predicted from cycle test	0.59 (0.51, 0.66)	0.67 (0.57, 0.75)		0.52 (0.43, 0.59)	0.58 (0.47, 0.68)	
Predicted from treadmill test	0.53 (0.44, 0.61)	0.69 (0.62, 0.74)	0.65 (0.54, 0.73)	0.44 (0.35, 0.53)	0.63 (0.56, 0.69)	0.59 (0.46, 0.68)
<i>DZ/sibling correlations</i>						
Measured	0.26 (0.11, 0.40)			0.43 (0.29, 0.55)		
Predicted from cycle test	0.45 (0.37, 0.53)	0.45 (0.34, 0.55)		0.45 (0.36, 0.54)	0.52 (0.41, 0.61)	
Predicted from treadmill test	0.43 (0.35, 0.51)	0.53 (0.45, 0.60)	0.37 (0.23, 0.49)	0.42 (0.32, 0.50)	0.53 (0.45, 0.61)	0.38 (0.24, 0.50)

CI, confidence interval.

Genetic Analyses

The twin and cross-twin/cross-trait correlations of measured $\dot{V}O_{2\max}$ and predicted $\dot{V}O_{2\max}$ are presented in Table 3. For $\dot{V}O_{2\max}$ in ml/min, MZ correlations ($r = 0.61$ for measured $\dot{V}O_{2\max}$ and $r = 0.67$ and $r = 0.65$ for the $\dot{V}O_{2\max}$ predicted from the submaximal cycle and treadmill tests) are almost twice as high as the DZ/sibling correlation ($r = 0.26$, $r = 0.45$ and $r = 0.37$). When the MZ resemblance is higher than the DZ resemblance this constitutes evidence for genetic influences on $\dot{V}O_{2\max}$. For $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹, twin correlations were also higher for MZ twins ($r = 0.53$, $r = 0.58$, and $r = 0.59$) than for DZ twins/siblings ($r = 0.43$, $r = 0.52$, and $r = 0.38$) but much less than half, providing evidence for genetic as well as shared environmental factors underlying familial aggregation. The cross-twin/cross-trait correlations (off-diagonal correlations in Table 3) are higher for MZ twins than for DZ twins/siblings for all phenotypes, suggesting genetic influences on the covariance between measured $\dot{V}O_{2\max}$ and $\dot{V}O_{2\max}$ predicted from the submaximal cycle and treadmill tests.

Genetic modeling started with an ACE model, as in all cases the DZ/sibling correlation was higher than half the MZ corre-

lation, except for measured $\dot{V}O_{2\max}$ in ml/min. Shared environmental influences were not significant for measured and predicted $\dot{V}O_{2\max}$ in ml/min [$\chi^2(6) = 10.8$, $P = 0.096$]. For $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹, shared environmental factors were not significant for the measured $\dot{V}O_{2\max}$, but for predicted $\dot{V}O_{2\max}$ a small but significant effect of shared environmental factors was detected. Standardized components from the best fitting model for additive genetic and shared and unique environmental influences on measured and predicted $\dot{V}O_{2\max}$ and their covariances are presented in Table 4. Heritability estimates for measured $\dot{V}O_{2\max}$ are 60% (95% CI: 47–69%) and 55% (95% CI: 43–64%) for $\dot{V}O_{2\max}$ in ml/min and ml·min⁻¹·kg⁻¹, respectively. Heritability estimates for predicted $\dot{V}O_{2\max}$ range from 47% for $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹ predicted from the treadmill test to 67% for $\dot{V}O_{2\max}$ in ml/min predicted from the cycle ergometer test. Shared environmental influences are small and not significant for $\dot{V}O_{2\max}$ in ml/min. For $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹, however, 12% (95% CI: 4–19%) of the variance in $\dot{V}O_{2\max}$ predicted from the cycle protocol and 4% (95% CI: 4–19%) of the variance in $\dot{V}O_{2\max}$ predicted from the treadmill protocol could be explained by shared environmental influences.

Table 4. Standardized estimates (95% CI) for additive genetic (A), shared environmental (C), and unique environmental (E) influences on measured $\dot{V}O_{2\max}$ and $\dot{V}O_{2\max}$ predicted from the submaximal cycle and treadmill tests and their covariances

	$\dot{V}O_{2\max}$, ml/min			$\dot{V}O_{2\max}$, ml·min ⁻¹ ·kg ⁻¹		
	Measured	Predicted from Cycle Test	Predicted from Treadmill Test	Measured	Predicted from Cycle Test	Predicted from Treadmill Test
<i>Additive genetics (A)</i>						
Measured	0.60 (0.47, 0.69)			0.55 (0.43, 0.64)		
Predicted from cycle test	0.76 (0.63, 0.85)	0.67 (0.60, 0.75)		0.70 (0.55, 0.82)	0.47 (0.32, 0.60)	
Predicted from treadmill test	0.70 (0.56, 0.81)	0.76 (0.65, 0.84)	0.64 (0.53, 0.72)	0.62 (0.46, 0.75)	0.61 (0.45, 0.75)	0.55 (0.42, 0.66)
<i>Shared environment (C)</i>						
Measured	—			—		
Predicted from cycle test	—	—		—	0.12 (0.04, 0.19)	
Predicted from treadmill test	—	—	—	—	0.09 (0.01, 0.16)	0.04 (0.00, 0.10)
<i>Unique environment (E)</i>						
Measured	0.40 (0.31, 0.55)			0.44 (0.35, 0.56)		
Predicted from cycle test	0.24 (0.15, 0.37)	0.33 (0.25, 0.43)		0.31 (0.19, 0.46)	0.41 (0.32, 0.52)	
Predicted from treadmill test	0.30 (0.19, 0.44)	0.24 (0.16, 0.35)	0.36 (0.28, 0.47)	0.37 (0.24, 0.54)	0.30 (0.21, 0.43)	0.40 (0.31, 0.52)

Heritability estimates in boldface. Dash indicates that this component could be constrained to be equal to zero.

Significant genetic correlations were found for measured $\dot{V}O_{2\max}$ and $\dot{V}O_{2\max}$ predicted from the submaximal cycle test [$r = 0.84$ (95% CI: 0.76–0.91) for $\dot{V}O_{2\max}$ in ml/min and 0.81 (95% CI: 0.68–0.95) for $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹]. Measured $\dot{V}O_{2\max}$ and $\dot{V}O_{2\max}$ predicted from the submaximal treadmill test show a genetic correlation of 0.73 (95% CI: 0.62–0.82) and 0.63 (95% CI: 0.48–0.76) for $\dot{V}O_{2\max}$ in ml/min and in ml·min⁻¹·kg⁻¹, respectively. A genetic correlation > 0 indicates that traits are influenced by common genes. Therefore, these correlations suggest that the three $\dot{V}O_{2\max}$ measures largely reflect the same set of underlying genetic variants. Furthermore, 61–76% of the phenotypic correlations between measured $\dot{V}O_{2\max}$ and $\dot{V}O_{2\max}$ predicted from the submaximal cycle and treadmill tests could be explained by genetic factors.

Meta-analysis

The literature search and screening resulted in 11 articles (see Table 1). Four studies were excluded from the meta-analysis. The studies by Montoye and Gayle (32), Lortie et al. (28), Lesage et al. (27), and Bouchard et al. (8) are parent-offspring studies and were excluded from the analysis because cohort effects and shared environment could be affecting the correlations. Moreover, whereas other studies either corrected for sex and age or used single-sex or age-restricted samples, Montoye and Gayle (32) and Lortie et al. (28) additionally corrected for skinfold thickness, physical activity, cigarette smoking, and social-economic status.

The seven studies included in the meta-analysis show MZ twin correlations for $\dot{V}O_{2\max}$ ranging from 0.62 to 0.95, whereas the DZ and sibling correlations are much lower (0.04 to 0.51). In the study by Sundet et al. (42), $\dot{V}O_{2\max}$ was predicted from extrapolation of the $\dot{V}O_2$ /HR slope, whereas the rest of the studies reported measured $\dot{V}O_{2\max}$ values. All of these remaining studies corrected the $\dot{V}O_{2\max}$ values for sex when the sample comprised both males and females, except for two studies by Klissouras and coworkers (24, 25). The age range in most studies was very restricted; two studies with a broader range corrected for age [Bouchard et al. (9) and the current study]. Univariate twin correlations (without siblings, estimated from a saturated model) from the current study were used in the meta-analysis ($r_{MZ} = 0.58$, $r_{DZ} = 0.29$, and $r_{MZ} = 0.54$, $r_{DZ} = 0.38$ for $\dot{V}O_{2\max}$ in ml/min and ml·min⁻¹·kg⁻¹, respectively).

Heterogeneity testing showed that all studies on the heritability of $\dot{V}O_{2\max}$ expressed in ml/min (combined sample of 1,088 individuals) could be taken together [$\chi^2(4) = 5.8$, $p = 0.218$], and a sample size weighted heritability estimate of 59% (95% CI: 52–66%) was found. For $\dot{V}O_{2\max}$ expressed in ml·min⁻¹·kg⁻¹ a weighted heritability estimate of 64% (95% CI: 60–69%) was found in a combined sample size of 3,120 individuals, but heterogeneity testing showed that these studies could not be simply taken together [$\chi^2(4) = 12.2$, $P = 0.016$]. Repeating the analysis without the study by Sundet et al. (42) (in which $\dot{V}O_{2\max}$ was predicted) removed heterogeneity in the estimates of the four remaining studies ($P = 0.098$) and increased the weighted heritability estimate to 72% ($n = 1,004$). For both $\dot{V}O_{2\max}$ expressed in ml/min and $\dot{V}O_{2\max}$ expressed in ml·min⁻¹·kg⁻¹, shared environmental influences were not significant ($P > 0.05$).

DISCUSSION

The main purpose of this paper was to estimate the heritability of aerobic fitness in an adolescent population, as assessed by $\dot{V}O_{2\max}$ measured during a maximal exercise test. In concordance with previous literature, $\dot{V}O_{2\max}$ was only moderately correlated with regular exercise behavior in leisure time. Genetic analysis revealed that 60% of the total variance in measured $\dot{V}O_{2\max}$ in ml/min and 55% of the total variance in measured $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹ can be explained by genetic factors.

In addition to measuring $\dot{V}O_{2\max}$ during the climax of a graded maximal cycle ergometer test, we predicted $\dot{V}O_{2\max}$ from submaximal tests on a cycle ergometer and a treadmill using extrapolation of the heart rate/oxygen uptake (HR/ $\dot{V}O_2$) curve to the predicted HR_{max}. Only a moderate phenotypic relationship was found between predicted $\dot{V}O_{2\max}$ and measured $\dot{V}O_{2\max}$ in the current study ($0.57 < r < 0.70$). This was lower than had been reported in previous studies of adult subjects (16, 19, 26). This difference can be attributed in part to the poor agreement between predicted and actual HR_{max}. Although there is substantial evidence that HR_{max} is age related in adults, it has been suggested that HR_{max} might be age independent in children and adolescents (36). When repeating the analysis using the measured HR_{max}, the phenotypic correlations between the measured $\dot{V}O_{2\max}$ and the predicted $\dot{V}O_{2\max}$ indeed increased. Nonetheless, correlations remain below those found for adults, suggesting that, apart from the higher individual variation in HR_{max}, the variability in the HR/ $\dot{V}O_2$ relationship may also be higher in adolescents.

Despite the moderate phenotypic correlation to measured $\dot{V}O_{2\max}$, heritability estimates from multivariate genetic analyses showed that heritability estimates for predicted $\dot{V}O_{2\max}$ (46–67%) were very similar to those obtained for measured $\dot{V}O_{2\max}$. For $\dot{V}O_{2\max}$ in ml/min, the heritability estimates were higher than measured $\dot{V}O_{2\max}$, but for $\dot{V}O_{2\max}$ in ml·min⁻¹·kg⁻¹ the heritability estimates were as high (treadmill test) or lower (cycle ergometer test) than measured $\dot{V}O_{2\max}$. However, as all heritability estimates are within the CI of measured $\dot{V}O_{2\max}$, the differences are not significant. Moreover, there was a substantial overlap in the genetic factors influencing predicted and measured $\dot{V}O_{2\max}$. That genetic effects on $\dot{V}O_{2\max}$ can be reliably estimated from submaximal tests is important as submaximal tests may be more suitable in large-scale studies. The graded maximal exercise test requires strenuous physical activity from the participant, producing discomfort, and cannot be attained by or poses a health risk for some subgroups of the population (e.g., sedentary individuals, young children, the elderly, or patients suffering from cardiovascular or respiratory disease). It may also lead to a larger selection bias when recruiting volunteers from population-based samples (like twin registries) as not all participants may be willing to exercise to exhaustion. This favors the participation of regular exercisers over sedentary subjects in exercise testing studies, which will lead to biased estimates of both mean and variance in $\dot{V}O_{2\max}$. The use of submaximal tests may lead to samples that are more representative of the general population.

Our sample size weighted meta-analysis on all heritability studies in children, adolescents, and young adults to date showed that 59% (when expressed in ml/min) ($n = 1,088$) and

72% (when expressed in $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$) ($n = 1,004$) of the variance in measured $\dot{V}O_{2\max}$ can be explained by genetic influences. All studies converge on the absence of detectable shared environmental factors (C). Shared environmental influences, including the family environment, were also low and not significant in the current study (except for predicted $\dot{V}O_{2\max}$ expressed in $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$), but the power to detect C was low, even after adding siblings of the twins to the design. Power analysis suggests that our sample size had to be at least twice as big for C to be detected with 80% power (35). This leads us to suggest that shared environmental influences on adolescent $\dot{V}O_{2\max}$ cannot be excluded but at best play a very modest role.

The overarching conclusion from our (meta-)analyses is that $\dot{V}O_{2\max}$ is a highly heritable phenotype from childhood to young adulthood. Heritability is likely to continue into adulthood, but there were no middle-aged or older adult twin samples that could be included in our meta-analysis. We did find four studies that measured $\dot{V}O_{2\max}$ in parents and offspring. In these parent-offspring designs, however, heritability estimation can be affected by cohort effects, since different genetic variants affecting aerobic fitness can be expressed at different ages. To get a complete picture of the heritability of $\dot{V}O_{2\max}$ across the whole life-span, twin studies focusing on middle-aged and older samples are direly needed.

A limitation of our study is that we cannot currently determine the exact contribution of the two different components that make up the heritability of $\dot{V}O_{2\max}$: genetic factors that contribute to baseline (untrained) performance levels and those related to “gain” in $\dot{V}O_{2\max}$ (i.e., genetic factors contributing to aerobic trainability). The HERITAGE study showed that the variation in baseline performance, as well as the variance in trainability, is larger between families than within families, confirming the role of genetic factors in baseline levels as well as in gain in $\dot{V}O_{2\max}$ (7, 8). Our study used a mixture of sedentary participants and moderately and vigorous exercisers. $\dot{V}O_{2\max}$ in the two latter groups will reflect a mixture of the baseline and trainability components. A possible way to discriminate between the two components is by estimating the heritability of $\dot{V}O_{2\max}$ in untrained (persistent sedentary) individuals only. A further limitation is that even though the current study is the largest twin study on measured $\dot{V}O_{2\max}$, our sample is still too small to have enough power to analyze sex differences in $\dot{V}O_{2\max}$. It might be that the effects of genetic or environmental factors on $\dot{V}O_{2\max}$ differ between males and females. A limitation of our meta-analysis is that there was significant heterogeneity across the studies, so that a single estimate therefore does not capture all individual studies adequately. However, recomputation of heritability in a restricted, more homogenous subset led to similar estimates. Finally, it should be noted that maximal exercise tests performed on a cycle ergometer generally yield lower $\dot{V}O_{2\max}$ values than maximal exercise tests performed on a treadmill due to a larger excising muscle mass. Comparing the heritability studies of $\dot{V}O_{2\max}$ performed on a treadmill and cycle ergometer showed that the two heritability estimates of treadmill-derived $\dot{V}O_{2\max}$ are slightly higher (24, 30), but these estimates are based on small sample sizes consisting of 10 yr olds. Replication of these studies in other age groups are needed to examine the effect of exercise equipment on the heritability of $\dot{V}O_{2\max}$.

Twin studies offer a unique opportunity to estimate the importance of genetic and environmental influences on a trait. Estimates of heritability inform us how much of the variation in a phenotype in a population sample is due to genetic variation and generally define the upper limit of the percentage of variance that is explained by genetics but do not reveal which and how many genes are involved. Therefore, an important next step is to identify the genetic variants underlying the heritability of $\dot{V}O_{2\max}$. Thus far we have seen case-control candidate gene and linkage studies, mostly characterized by small sample sizes and mixed results (11). Two of the most studied polymorphisms are the R577X variation in the *ACTN3* gene and the I/D polymorphism in the *ACE* gene (29, 40). The preferred approach to identify genetic variants for complex traits (which are known to be influenced by multiple genetic factors) is a meta-analysis of genome-wide association (GWA) studies with a large cumulative sample size (18, 45). However, only one GWA study on $\dot{V}O_{2\max}$ has been conducted to date by Bouchard et al. (12). Strikingly, despite the small sample size, this study revealed that 16 single nucleotide polymorphisms (SNPs) accounted for 45% of the variance in gains in $\dot{V}O_{2\max}$ after exposure to a standardized 20 wk exercise program in a sample of 473 sedentary adults (12). No GWA studies have yet been performed on $\dot{V}O_{2\max}$ in the untrained or baseline state (before training). Such studies will need large samples with both $\dot{V}O_{2\max}$ data and genome-wide genotyping. The feasibility of this increases greatly if submaximal exercise tests generate sufficiently valid estimates. Notwithstanding the imperfect correlation between predicted and measured $\dot{V}O_{2\max}$, our results can be considered encouraging: The high genetic correlation between measured and predicted $\dot{V}O_{2\max}$ in the current study suggests that they largely capture the same latent genetic factors and these genetic factors explained the largest part of the observed correlation between measured and predicted $\dot{V}O_{2\max}$. GWA meta-analyses across studies using (graded) submaximal and maximal tests should be able to pick up these shared genetic variants.

To conclude, the results of the current study, together with the results of the meta-analyses, confirm that innate factors determine more than half of the individual differences in the $\dot{V}O_{2\max}$ from childhood to young adulthood.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

AUTHOR CONTRIBUTIONS

Author contributions: N.M.S., I.N., M.B., and E.J.d.G. conception and design of research; N.M.S. and I.N. performed experiments; N.M.S. and E.J.d.G. analyzed data; N.M.S., I.N., M.B., and E.J.d.G. interpreted results of experiments; N.M.S. prepared figures; N.M.S. drafted manuscript; N.M.S., I.N., J.J.H., M.B., and E.J.d.G. edited and revised manuscript; N.M.S., I.N., J.J.H., M.B., and E.J.d.G. approved final version of manuscript.

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